



Moderation of water deficit stress-induced damages by the application of silver nano particles and indole butyric acid foliar spray in maize

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Abstract

Drought is a persistent and intricate natural vulnerability whose rate of recurrence and enormity is expected to increase with climate change. Regardless of progress in response and adaptation to water scarcity (with the development of new policies), drought continues to cause severe impacts and affliction. Therefore, the present findings have focused on the calculation of the impact of silver nanoparticles (Asp-AgNP) and indole butyric acid (IBA) on the physiological mechanism of water deficiency tolerance in a special maize variety, i.e Sarhad yellow under induction of 7 and 10 days of drought at vegetative stage. The maize seeds (Sarhad yellow) of the CCRI, Persabaq Nowshera were collected and sown in triplicate in specific diameter clay pots, in an experimental house located in the botanical department of the University of Peshawar. The nanoparticles were analyzed by scanning electron microscopy (SEM), the dispersive energy X-ray spectroscopy (EDX) confirmed the size of the 73–166nm nanoparticles with thick surfaces. TGA (thermogravimetric analysis) and differential thermal analysis (DTA) showed a significant reduction in endothermic mass and a greater exothermic appearance. Drought stress has reduced the agronomic and physiological characters counting the content of chlorophyll "a", "b", carotenoids and proteins together with peroxidase (POD). Asp-AgNP was more effective with a minimum of 7 days of drought, which improves the physiological attribute alone and in combination with IBA compared to the condition of maximum drought stress. It is hereby accomplished that the inhibitory effect of short-term water deficit stress has been improved by the exogenous application of Asp-AgNP and IBA foliar spray, but yield loss via long term osmotic stress will not be encountered using growth regulators and nanoparticles. Climate smart agriculture is the only way to overcome the problems created by the long-term exposure of cereal crops to climate change, including water deficit stress.

Keywords: Maize, Drought, Nanoparticles, Nanotechnologies, Osmolites, Antioxidants

1. Introduction

One of the most important cereal crops is maize (*Zea mays* L.), which is found everywhere, both in rainy regions and in arid areas. It is taking on a unique position all over the world, especially in Pakistani agriculture due to its greater capacity to yield in a shorter period of time. Maize is a rich source of food and forage. It is used in the production of corn oil, corn jams, corn sugar, corn protein and syrup, etc. in the industry ^[1]. The production of (*Zea mays* L.) is strongly influenced by varying climatic conditions in different areas of the world in different ways ^[2]. Late or early sowing of maize greatly influences the production and growth of crops due to changes in the climatic conditions of a region. However, such variations in climatic conditions influence the development and production differently, depending on the immensity of the change. Considerable effects on maize development and production have already been recorded by changing the climate ^[3, 4]. Drought is considered as a cosmopolitan crisis, dangerous for the growth of crops and consequently, for the conservation of shelters ^[5]. Numerous studies have developed different drought indices to estimate the severity of drought due to a significant reduction in water availability. Water deficit stress is a terrifying danger for our agriculture that cannot be avoided. Kramer (1980) estimated that 33% of arable land in the world faces drought, which is the main cause of crop productivity reduction ^[6]. The severity of water deficit stress cannot be predicted because it is controlled by many factors, namely the presence and distribution of precipitation, the

soil's ability to accumulate moisture and the demand for evaporation ^[7]. In conclusion, water scarcity damages different physiological and metabolic processes in plants. Drought results in retarded growth, a reduction in chlorophyll pigments and changes in fluorescence parameters ^[8, 9]. Nanotechnology is considered as the science of manipulating objects at the nanoscale (up to 1 / 100,000 of the width of a human hair) to generate things and fresh goods with great potential to change society. It offers an escalating analysis in many fields, such as electronics, medicines and life sciences ^[10, 11]. NPs are molecular aggregates that have at least a size between 1 and 100 nm ^[12, 13], with the ability to severely alter their physico-chemical properties with respect to bulk material ^[14]. Nanoparticles can be prepared from a complete variety of bulk substances and can expand their behavior depending on the chemical composition and size/shape of the particles ^[15]. Nanoparticles can be synthesized through the use of numerous metallic or non-metallic elements that have distinctive properties and widespread applications in various fields of science and medicine ^[16]. By considering the changes in climate expected in the future, this study is aimed to investigate the effects of climatic variation in terms of providing water deficit condition as induced drought stress to the maize crop to examine their growth responses, osmo-protection capabilities and their antioxidant enzyme system under the effect of IBA foliar spray and Silver nanoparticles. This study is of high importance because Pakistan is in threat of water scarcity because of global

variation in terms of climate change and the food security situation across the country is not very good as it is estimated that with rise of temperature (0.5°C–2°C), agricultural productivity in Pakistan will decrease up to 8%–10% by 2040. To stabilize our agricultural growth rate Pakistan needs to put in place immediate adaptive measures recommended by the present study as well as related findings of other researchers.

2. Materials and methods

2.1 Preparation of Asp-AgNPs nanoparticles

Glucose, aspartate, sodium hydroxide, hydrochloric acid etc were acquired from Sigma-Aldrich. Then we prepared the silver nanoparticles using the reduction method. We dissolved 18 mg of glucose in 100 ml of deionized water to obtain a concentration of 1.0 mM. We prepared a 1.0 mM solution of AgNO₃ by mixing 16.99 mg of AgNO₃ with 100 ml of deionized water to which we added the aspartate solution (13.3 mg / 100 ml) drop by drop. Then we added the glucose solution to the aspartate and to the silver nitrate solution drop by drop, constantly shaking it with a magnetic stirrer. The precipitates formed were centrifuged at 3000 rpm for 20 minutes at 250 ° C. Then it was dried during the night. Then we stored the resulting powders in closed vials in environmental circumstances.

2.2 Analysis of Asp-AgNPs nanoparticles. The Asp-AgNPs were exposed to morphological characteristics using SEM. The prepared Asp-AgNPs were filmed with carbon tape and exposed to gold coating using Spi-module, sputter coater and examined morphological features through FE-SEM (JSM-5910-JEOL-JAPAN). The Asp-AgNPs were characterized for elemental analysis by X-ray spectroscopy (EDS). We then performed thermogravimetric analysis using TA, Pyris Diamond Series TG / DTA instruments manufactured by Perkin Elmer, USA under an air atmosphere included in the temperature range 300 ° C - 6000 ° C at a heating rate of 100 ° C / min, using ceramic (Al₂O₃). A sample of 6,446 mg was initially heated to 50°C for 1.0 minutes to remove the adsorbed solvent. We then heated the sample from 600 °C to 10 °C/min and kept it at 600 °C for 20 minutes. Then we measured the weight loss between the weight measured at 300 ° C and 6000 ° C.

2.3 Pot experiment

Seeds of *Zea mays* L local accessions sarhad yellow were obtained from CCRI Nowshera, Pakistan. The seed were sown in triplicates in clay pots having the lower diameter of 18cm and upper inside diameter, 20cm height and 2cm thickness) filled with 2.0kg of dried soil and silt with a ratio of 2:1 having EC 2.85-6.70, pH 6.8-7.3 and moisture content 15-20% arranged in complete randomized design in experimental house of Botany department University of Peshawar. When the seeds were fully germinated, the plants were exposed to drought stress of 7days and 10days along with untreated control whereas; Asp-AgNPs and IBA foliar spray were used in the designed experiment. Standard practices satisfactory for pot experiment were followed and the experiment was kept far away from the reach of pest and other diseases. The plants were then watered according to the requirement.

2.4 Agronomic analysis

2.4.1 Germination percentage

After 7 days of seed sowing, we noted percentage of seeds germinated by following the method of [17].

2.4.2 Shoot and root parameter

After the date of germination, root and shoot length (cm), fresh and dry weight of root and shoot, leaf area (cm²) and root/shoot ratio was noted.

2.4.3 Seedling vigorous index (SVI)

Seedling vigorous index was noted by following Abdul-Baki and Anderson's process. The SVI was demonstrated in number of means and standard deviation.

Eq.1. SVI = the length of shoot + root × percentage of germination.

2.5 Physiological and biochemical attributes

2.5.1 Calculation of soluble protein content (SPC)

Protein content was studied using recommended procedure of [18]. Using mortar and pestle, we grinded the fresh leaves with a weight of 0.2g in phosphate buffer with pH 7.5. Then we poured 0.1 ml of the extract over into the tubes and a total volume of 1 ml was made with distilled water. Then we added 3.0 ml reagent which had (3g of sodium carbonate (Na₂CO₃) 0.6g of NaOH (0.1 N) and 1.5 g of Na-K tartrate dissolved in 150 ml of distilled water and CuSO₄.5H₂O (0.125 g) dissolved in 25 ml of water distillate). By shaking for 10 minutes, 0.1 ml of Foline phenol reagent was added. The absorbance of each sample was observed at 650 nm after 30 minutes of incubation.

2.5.2 Determination of soluble sugar content (SSC)

In order to determine the soluble sugar content, 0.5g material was added to 10ml of distilled water. After mixing it fully, it was then centrifuged at 3000 rpm for 5 min. 1ml of 80% (w/v) phenol was mixed with 0.1ml of supernatant followed by the addition of 3.0ml concentrated sulphuric acid after room temperature incubation for 4 hrs then recorded at 420 nm using spectrophotometer.

2.5.3 Calculation of total chlorophyll content (TCC)

The procedure of Aron [19] was followed for evaluation of total chlorophyll content at absorbance of 645nm for chlorophyll "a" and 663nm for chlorophyll "b" in the selected foliar material by crushing 0.5g material in 80% acetone (cold).

2.5.4 Determination of total proline content (TPC).

The procedure of Bates [20] was used to estimate proline in foliar material. We crushed the plant materials of 0.5g in 10ml of 3% aqueous sulphosalicylic acid which was then subjected to filtration. By reacting 2ml of filtrate with 2ml acid ninhydrin 2ml of glacial acetic acid in a test tube for one hour at 100°C, the reaction was terminated in ice bath. The reaction mixture was extracted with 4ml toluene. The chromophore containing toluene was aspirated from the aqueous phase, warm to room temperature and absorbance read at 520nm against toluene as blank.

2.5.5 Determination of POD activity.

The POD content estimation was performed using the standard protocol [21]. The peroxidase activity was calculated

following the test mixture which had 0.1 ml of enzyme extract, 1.35 ml of 100 mM MES buffer (pH 5.5), 0.05% of H₂O₂ and 0.1 % phenylene diamine. The change in absorbance was recorded at 485 nm for 3 minutes with a spectrophotometer. POD activity was presented as ΔOD 485 nm/min mg of protein.

2.5.6 Determination of SOD activity.

The SOD content was estimated using the established protocol of [22]. The reaction combination (3 ml) was composed of 13 mM of methionine, 0.075 mM NBT, 0.1 mM of EDTA, 0.002 mM of riboflavin and 0.1 ml of enzyme extract in 50 mM phosphate buffer (pH 7.8). The mixture in the tube was kept under the light chamber for about 15 minutes. The absorbance was read at 560 nm with a spectrophotometer.

2.6 Statistical Analysis.

Complete research was designed in triplicates. Outcomes are expressed as mean \pm standard whereas; significance different at $P \leq 0.05$ and means relationship were done by following one-way ANOVA using STASTICA 8.1.

3. Results

3.1 Physical Characterizations of Asp-AgNPs.

The EDX spectrum of silver nanoparticles showed strong 3.0keV signal peaks; while the peaks were at 20.194keV junction energies. Other peaks are C and O. No other impurity peaks have been observed. This shows that the silver nanoparticle model contains pure silver as shown in Figure 5a. The SEM image of Asp-AgNPs showed that almost all Asp-AgNPs have a spherical shape. The size of silver nanoparticles varies from 73nm to 166nm and their average size is 113.52nm, as shown in Figure 9 (b-c). While looking at the TGA curve, it is clear that no loss of weight of the sample occurred at temperatures between 50 ° C and 600 ° C. Therefore, it is thermally stable. The DTA diagram showed a strong exothermic peak, the main peak was observed at 250 ° C which is mainly attributed to the crystallization of Asp-AgNP simultaneously, as shown in Figure 10 (a-c).

3.2 Agronomic and Physiological Study.

Significant amount of reduction was observed in agronomic characters including root/shoot ratio, leaf area, seedling vigorous index (SVI) and germination percentage on the medium in the chosen cultivar (Table 1) having greater detrimental effects on T₁ (7 days drought), T₃ (7 days drought + IBA) and T₈ (10 days drought + NPs + IBA). Foliar spray of Silver nanoparticles and Indole Butyric acid controlled the effects of drought stress upto some extent on the above-mentioned attributes, although in the T₁₀ (untreated + NPs), T₁₁ (untreated+IBA) and T₁₂ (untreated + NPs + IBA) reported maximum at the ambient drought stress, whereas the moderate amplitude has been reported in the T₂ (7 days drought + NPs), T₄ (7 days drought + NPs + IBA), T₆ (10 days drought + NPs) and T₇ (10 days drought + IBA). By looking in to our consequences we can easily say that Silver nanoparticles and Indole butyric acid (growth regulator) can help the maize crop to show some resistant against the stress produced by the deficiency of water in terms of agronomic characters however it will not be that much effective against the long term exposure.

Drought stress also reduced the chlorophyll "a", chlorophyll

"b", total chlorophyll and SOD content in the medium in the selected cultivar (at $P \leq 0.05$ Fig 1,2 and 7) with the minimum amplitude of chlorophyll "a" in T₂ (7 days drought + NPs). Foliar spray of Silver nanoparticles and Indole Butyric acid controlled the damaging effects of drought stress. The highest magnitude was reported in T₅ (10 days drought) and T₁₀ (untreated + NPs) whereas the moderate amplitude has been reported in T₁ (7 days drought), T₃ (7 days drought + IBA), T₄ (7 days drought + NPs + IBA), T₈ (10 days drought + NPs + IBA) and T₉ (untreated). The water deficiency has also damaged the proteins, proline, carotenoid contents, sugar and POD contents with the decreased chlorophyll a/b ratio in the choosen maize variety. The individual and combined effects of Silver nanoparticles and IBA has ameliorated the detrimental effects of water dificit stress on above-mentioned attributes, T₂ (7 days drought + NPs), T₄ (7 days drought + NPs + IBA) and T₁₀ (untreated + NPs) reported maximum at $P \leq 0.05$, whereas the greater negative effects were observed in T₃ (7 days drought + IBA), T₅ (10 days drought) and T₇ (10 days drought + IBA), although the moderate amount of the above mentioned attributes were found in T₁ (7 days drought), T₈ (10 days drought + NPs + IBA), T₉ (untreated), T₁₁ (untreated + IBA) and T₁₂ (untreated + NPs + IBA).

4. Discussion

Drought is an intense climatic affair that is very dangerous because it develops slowly and often sneaks up on one. The intensified drought condition especially with respect to its duration can have major consequences, making it one of the deadliest natural hazards. Though by applying soil amendments, one may be able to alleviate the hazardous effects of drought stress. To confirm this statement, the current research was carried out to observe the impacts of Silver nanoparticles alone and in combination with indole butyric acid on maize growth, under drought stress. The water deficiency significantly minimized the morpho-physiological growth of maize. Enough water availability is necessary for best growth, productivity and floristic variations. Therefore the production and growth of plants might be influenced by the induced changes in physiological processes and agronomic characters under drought. The current examination revealed that water deficit stress sensitivity or tolerance of selected maize variety may not only vary in physiological processes such as osmolytes, phyto-hormonal activities but it also changes in the protein content. Amplified crop yield is one of the major needs of forthcoming population growth, but drought stress has the tendency to minimize the crop yield. Result in terms of EDX spectrum of Silver Nanoparticles showed mighty signal peaks at 3.0keV; while peaks were built at the binding energies of 20.194 keV (Fig 5a). Our result is in harmony with previous work reported the stable AgNPs synthesized from extract of *Allium cepa* the silver absorption peak at 3 keV confirmed AgNPs synthesis [23]. In TGA curve (Fig 6a) no mass dissipation in model was observed in temperature between 50°C-600°C, thus it is thermally stable. DTA showed (Fig 6b) major peak at 250°C which mainly featured to crystallization of Asp-AgNps. Using TGA, various groups have ameliorated thermal stability in nanotube/polymer composites in comparison to neat polymers [24]. Present findings revealed that agronomic character of maize as shown in (Table 1) significantly

declined with the enlargement in the amplitude of drought stress at vegetative stages in selected variety. Our findings are similar to [25] that depletion of root length under water deficiency may be because of obstruction of cell division and elongation leading kinds of tuberization. This fall up in growth is due to low osmotic potential as well as declination in wall extensibility and cellular expansion. Drought stress for 12 days retarded the growth and reduced the dry weight of maize, but the plants recovered after watering for 6 days [26]. Water deficit stress also reduced the shoot fresh weight and of shoot in maize [27, 28]. Results of our study displayed the significant variation in all physiological and biochemical measure in the selected variety (sarhad yellow) of maize at

vegetative stage. Short term exposure to drought caused increase in carotenoids, proteins, proline, sugar and POD content whereas chl “a” and “b”, total chlorophyll and SOD content was reduced (Fig 1-4). Our result is in line with previous work indicating that dryness of soil influence the chlorophyll “a” and “b” activity [29]. The declination in pigment content was showed in sensitive maize cultivar under water deficit stress [30]. Similar findings have been documented that Si is able to reduce the amount of proline in plants under water deficiency [31]. In reply to drought, proline accumulation normally occurs in cytosol where it plays a huge role in cytoplasmic osmotic adjustment [32].

5. Tables and figures

Table 1: Effect of Asp-AgNPs and Indole butyric acid foliar spray on root length, root fresh weight, root dry weight and % moisture content of maize (*Zea mays L*) under drought stress

Treatment	Root Length	Root Fresh Weight	Root Dry Weight	% Moisture Content
T1	6.53±1.15	0.566±0.059	0.173±0.012	69.2±1.45
T2	7±1.47196	0.62±0.2531	0.163±0.049	72.6±2.58
T3	5.96±0.47	0.476±0.091	0.17±0.0244	63.8±2.99
T4	7.56±2.51	0.38±0.1496	0.126±0.062	68.0±3.65
T5	6.53±1.06	0.34±0.0864	0.093±0.026	72.5±3.08
T6	6.26±1.55	0.286±0.102	0.09±0.0326	68.6±0.28
T7	6.66±0.54	0.423±0.148	0.12±0.0294	70.5±3.84
T8	5.06±1.47	0.353±0.101	0.106±0.041	62.7±25.1
T9	7.46±0.53	0.383±0.098	0.12±0.0454	69.6±3.84
T10	8.4±0.993	0.573±0.217	0.11±0.0408	80.5±1.08
T11	9.83±3.38	0.58±0.1143	0.09±0.0294	84.4±3.66
T12	7.83±0.97	0.706±0.089	0.183±0.053	73.5±7.85

T1=7 Days drought, T2=7DD+NPs, T3=7DD+IBA, T4=7DD+NPs+IBA, T5=10Days drought, T6=10DD+NPs, T7=10DD+IBA, T8=10DD+NPs+IBA, T9=Untreated, T10=Untreated+NPs, T11=Untreated+IBA, T12=Untreated+NPs+IBA.

Table 2: Effect of Asp-AgNPs and Indole butyric acid foliar spray on shoot length, shoot fresh weight, shoot dry weight and % moisture content of maize (*Zea mays L.*) under drought stress

Treatment	Shoot Length	Shoot Fresh Weight	Shoot Dry Weight	%Moisture Content
T1	29.9±2.71	2.56±0.62	0.293±0.087	88.7±0.98
T2	45.4±6.73	4.53±0.84	0.576±0.123	87.3±0.87
T3	35.9±7.67	3.42±1.45	0.483±0.180	85.3±1.23
T4	39.2±3.23	2.94±0.63	0.406±0.089	86.2±0.17
T5	31.6±2.80	2.5±0.208	0.32±0.0424	87.2±0.64
T6	40.4±6.56	3.50±0.46	0.433±0.108	87.7±1.55
T7	41.6±11.3	3.89±2.60	0.52±0.3536	86.6±0.46
T8	36.1±6.08	2.90±0.45	0.406±0.081	86.0±0.90
T9	47.8±3.80	3.72±0.88	0.563±0.164	85.0±1.96
T10	53.8±14.9	7.22±3.76	0.803±0.286	87.4±2.82
T11	56.5±3.51	6.04±0.72	0.71±0.1444	88.3±1.10
T12	64.3±3.84	8.54±1.90	1.216±0.158	85.4±1.47

T1=7 Days drought, T2=7DD+NPs, T3=7DD+IBA, T4=7DD+NPs+IBA, T5=10Days drought, T6=10DD+NPs, T7=10DD+IBA, T8=10DD+NPs+IBA, T9=Untreated, T10=Untreated+NPs, T11=Untreated+IBA, T12=Untreated+NPs+IBA.

Table 3: Effect of Asp-AgNPs and Indole butyric acid foliar spray on Leaf length, Leaf fresh weight, Leaf dry weight and % moisture content of maize (*Zea mays L.*) under drought stress

Treatment	Leaf Length	Leaf Width	Leaf Fresh Weight	Leaf Dry Weight	% Moisture Content
T1	7.83±2.54	1.8±0.216	0.516±0.154	0.066±0.020	87.12±0.300
T2	9.41±4.12	1.73±0.23	0.896±0.140	0.126±0.016	85.78±1.145
T3	7.33±4.30	1.63±0.38	0.693±0.282	0.103±0.036	84.70±1.006
T4	8.74±1.75	1.36±0.30	0.606±0.156	0.086±0.020	85.61±0.432

T5	8.73±2.36	1.46±0.12	0.573±0.659	0.08±0.0081	86.02±0.265
T6	11.3±4.57	1.8±0.081	0.756±0.152	0.113±0.032	85.25±1.340
T7	8.01±7.78	1.6±0.432	0.773±0.436	0.116±0.065	84.89±0.662
T8	7.03±4.50	1.33±0.23	0.56±0.1667	0.09±0.0216	83.66±0.943
T9	14.4±2.36	1.53±0.12	0.873±0.172	0.143±0.037	83.79±2.071
T10	15.1±8.24	1.96±0.44	1.21±0.4327	0.12±0.0725	90.85±2.474
T11	13.3±3.64	1.83±0.23	1.143±0.151	0.136±0.024	87.70±3.098
T12	16.3±7.05	2.46±0.04	1.576±0.457	0.143±0.061	90.00±5.909

T1=7 Days drought, T2=7DD+NPs, T3=7DD+IBA, T4=7DD+NPs+IBA, T5=10Days drought, T6=10DD+NPs, T7=10DD+IBA, T8=10DD+NPs+IBA, T9=Untreated, T10=Untreated+NPs, T11=Untreated+IBA, T12=Untreated+NPs+IBA.

Table 4: Effect of Asp-AgNPs and Indole butyric acid foliar spray on Root/Shoot, Leaf area, SVI and Germination % of maize (*Zea mays* L.) under drought stress

Treatment	Root/Shoot	Leaf Area	SVI	Germination %
T1	0.651±0.20	28.0±6.17	2798.3±4.83	0.766±1.24
T2	0.277±0.02	37.5±7.94	3320.7±3.86	0.633±0.47
T3	0.399±0.12	29.2±10.8	2376.2±7.68	0.566±0.94
T4	0.292±0.08	26.0±6.51	4212.0±4.68	0.901±0.81
T5	0.302±0.10	23.7±2.75	2796.4±4.83	0.733±1.24
T6	0.201±0.03	33.3±5.27	3269±6.628	0.701±0.81
T7	0.310±0.14	34.1±18.3	2737.0±20.2	0.566±1.69
T8	0.273±0.13	23.2±6.43	2058.3±12.3	0.502±1.63
T9	0.212±0.04	41.3±5.44	3131.7±5.40	0.566±1.24
T10	0.13±0.003	46.3±21.2	3732.0±13.0	0.601±0.81
T11	0.128±0.03	47.0±10.0	3760.7±3.25	0.566±0.47
T12	0.147±0.02	65.4±10.3	3850.6±6.00	0.533±1.24

T1=7 Days drought, T2=7DD+NPs, T3=7DD+IBA, T4=7DD+NPs+IBA, T5=10Days drought, T6=10DD+NPs, T7=10DD+IBA, T8=10DD+NPs+IBA, T9=Untreated, T10=Untreated+NPs, T11=Untreated+IBA, T12=Untreated+NPs+IBA.

Table 5: EDX result showing percentage of elements in Asp-AgNPs

>	Weight%	Atomic%
C K	2.52	15.57
O K	4.41	20.46
Ag L	93.07	63.98
Totals	100.00	-----

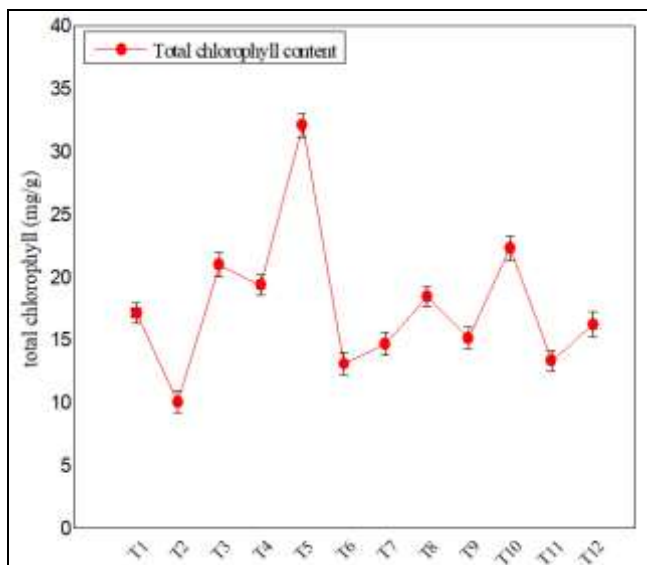


Fig 1: Effect of Asp-AgNPs and indole butyric acid foliar spray on chlorophyll “a” and “b” content of maize (*Zea mays* L.) Under (*Zea mays* L.) under drought drought stress

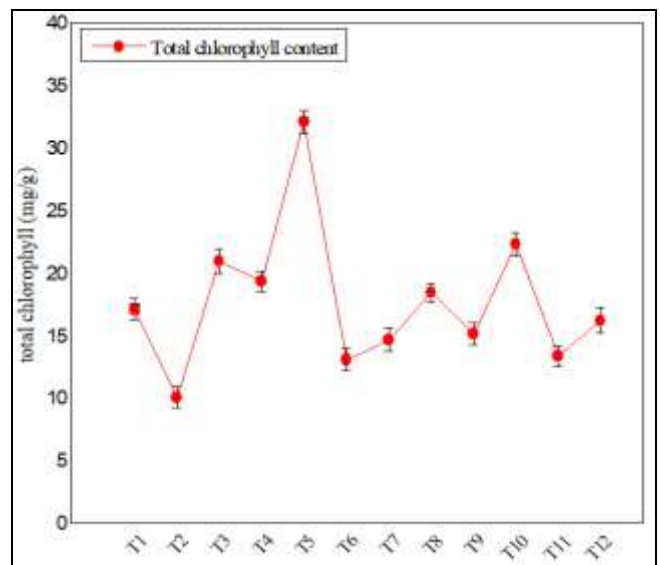


Fig 2: Fig 2.Effect of Asp-AgNPs and indole total chlorophyll content of maize stress

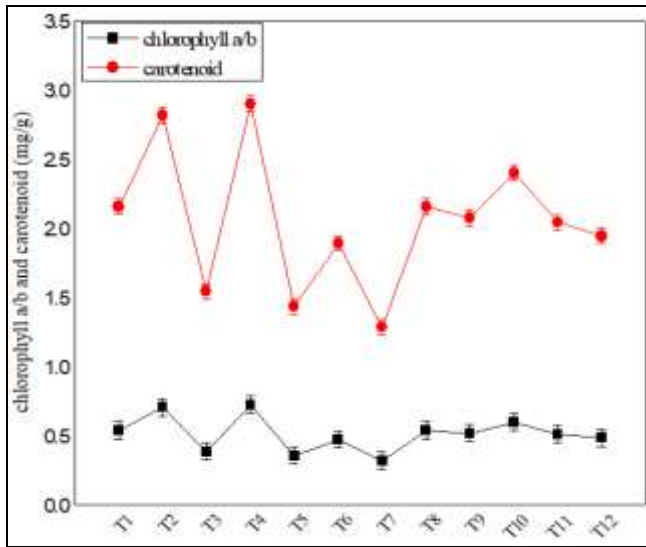


Fig 3: Effect of Asp-AgNps and indole butyric acid foliar spray on chlorophyll a/b and carotenoid content of *maize mays L.* Under drought stress (*Zea mays L.*) under drought stress

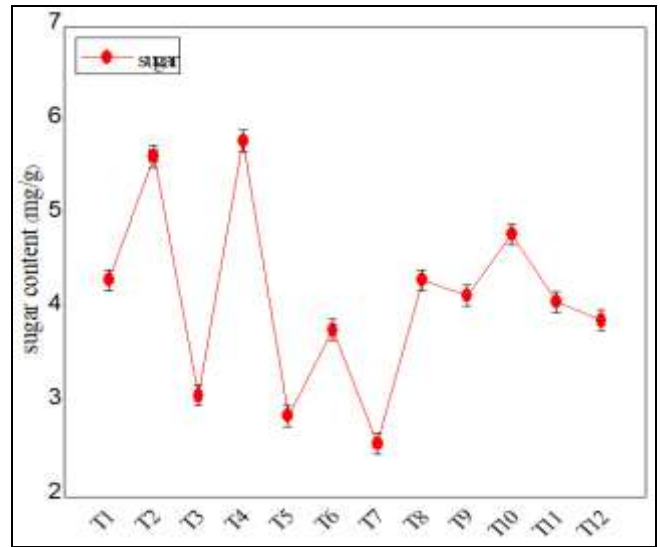


Fig 6: Effect of Asp-AgNps and on SOD content of maize (*Zea*

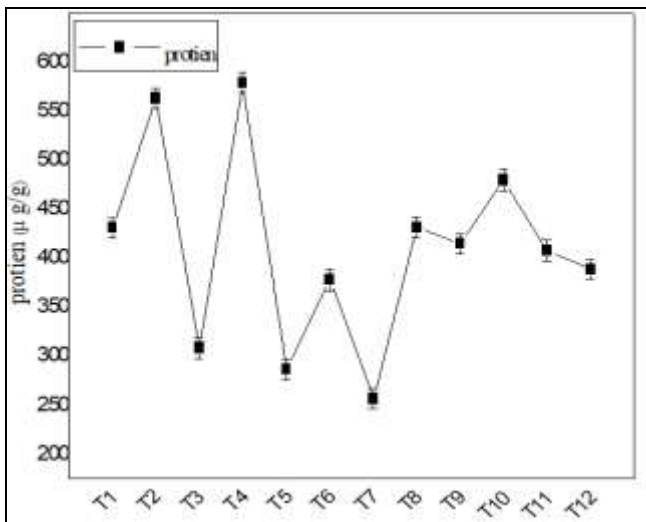


Fig 4: Effect of Asp-AgNps and indole protein content of maize (*Zea*

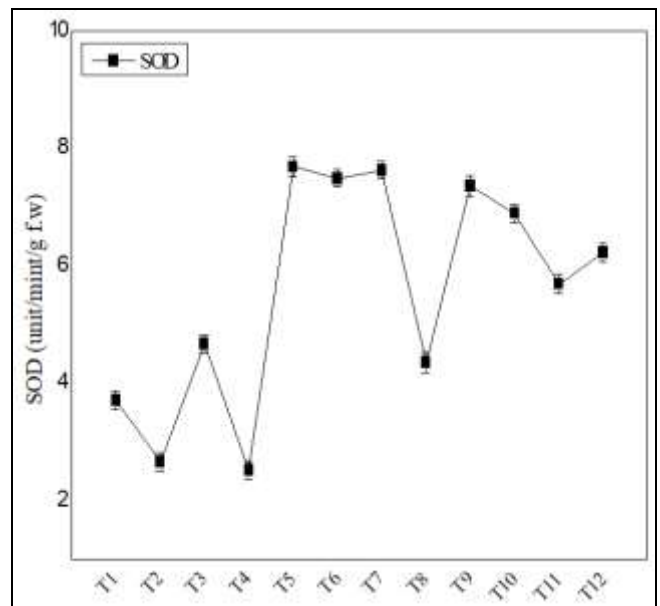


Fig 7: Effect of Asp-AgNps and indole butyric acid foliar spray indole butyric acid foliar spray on POD content of maize (*Zea mays L.*) under drought stress (*mays L.*) Under drought stress

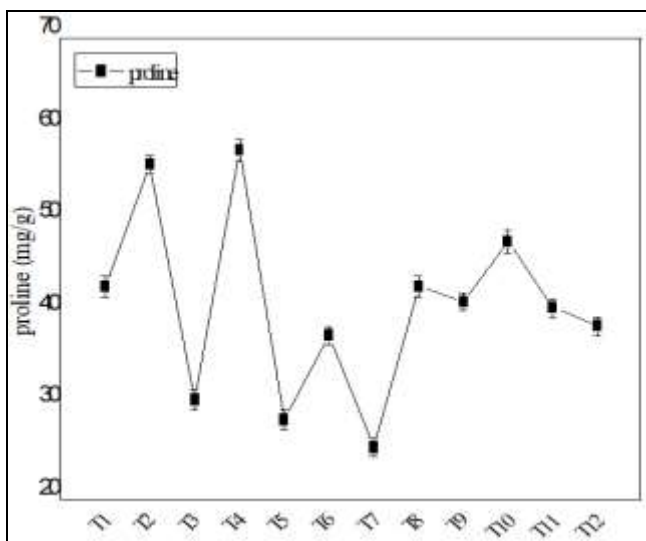


Fig 5: Effect of Asp-AgNps and indole butyric acid foliar spray indole butyric acid foliar spray on proline content of maize (*Zea mays L.*) under drought stress (*mays L.*) Under drought stress.

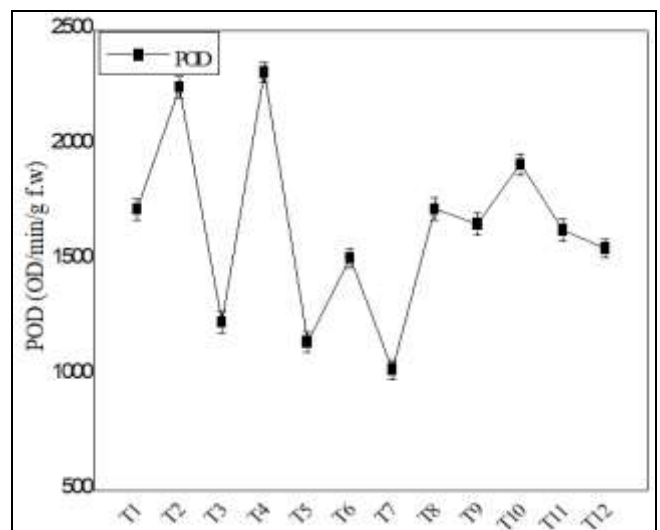
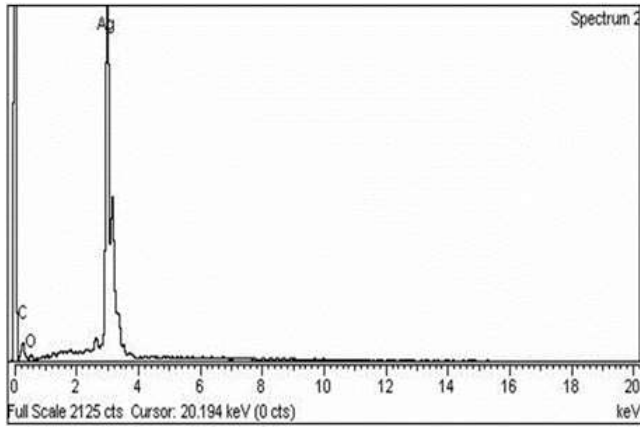
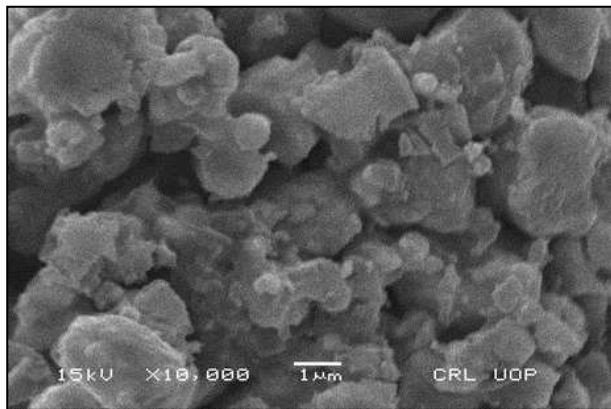


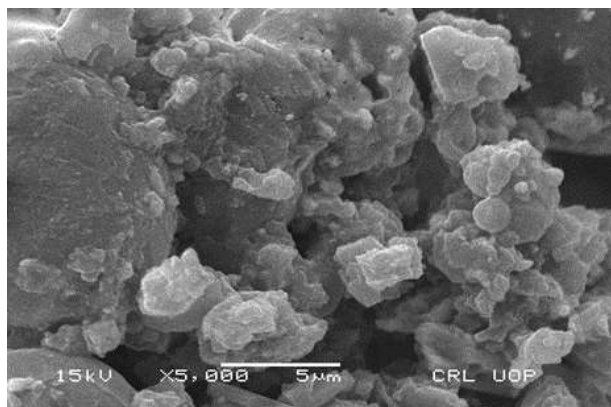
Fig 8: Effect of Asp-AgNps and on SOD content of maize (*Zea*



(a)

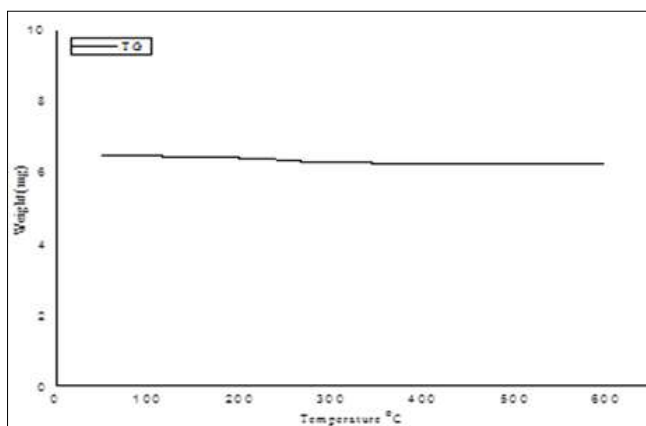


(b)

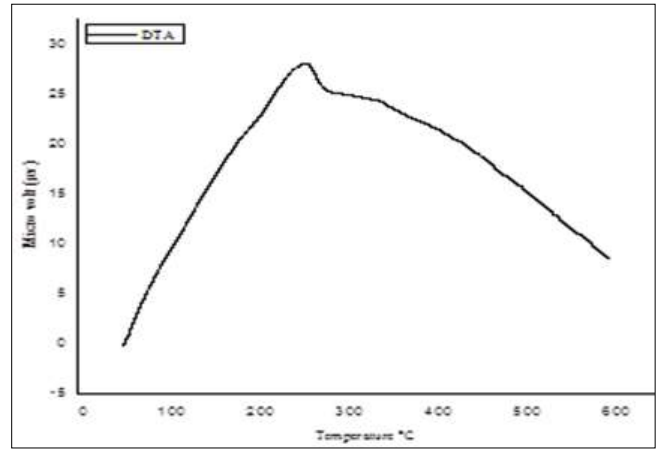


(c)

Fig.9 Characterization of Asp-AgNPs (a) Energy Dispersive X-ray Spectroscopy (EDX) of Asp-AgNPs (b-c) Scanning Electron Microscopy of Asp-AgNPs



(a)



(b)

Element	Weight%	Atomic%
C K	2.52	15.57
O K	4.41	20.46
Ag L	93.07	63.98
Totals	100.00	

(c)

Fig 10: Characterization of Asp-AgNPs (a) Thermogravimetric Analysis (TG) of Asp-AgNPs (b) Differential Thermal Analysis (DTA) of Asp-AgNPs (c) EDX results showing percentage of elements in Asp-AgNPs

5. Conclusion

After all our efforts we came to the conclusion that amplified water deficit stress have hazardous effects on the development of selected variety of maize (sarhad yellow) including physiological attributes, while the lower limit of 7 days drought stress showed comparatively less-harmful effects on physiological and agronomical attributes and been ameliorated by applying silver nanoparticles as an individual treatment and in association with IBA. These findings could have practical implications in the future to ensure agricultural, economic and environmental benefits. However, future field studies are needed to clarify the effects of nanoparticle changes on soil health and crop productivity at the molecular level.

6. References

1. Minfal. Statistics of Pakistan, Ministry of Food, Agric. & Livest. Econ. Wing, Islamabad, Pakistan, 2009.
2. Wittmer M, Auerswald K, Tungalag R, Bai YF, Schaeufele R, Bai CH. *et al.* Carbon isotopes discrimination of C3 vegetation in central Asian grassland as related to long-term and short-term precipitation patterns. *Bio geoscience Discussions.* 2008; (5):903-935.
3. Binder JS, Link GJ, Claupein W, Dai MLM, Wang P. Model based approach to quantify production potentials of summer maize and spring maize in the North China Plains. *Agron. J.* 2008; 100:862-873.
4. Meza FJ, Silva D, Vigil H. Climate change impacts on irrigated maize in Mediterranean climates: Evolution of double cropping as an adaptation alternative. *Agric. Systems.* 2008; 98:21-30.
5. Jaleel CA, Manivannan P, Wahid A, Farooq M, Somasundaram R, Paneerselvam R. *et al* Drought stress in plants: a review on morphological characteristics and pigments composition. *Int. J Agric. Biol.* 2009; 11:100-105.

6. Kramer PJ. Drought stress and origin of adaptation. In N.C. Turner, and P.J. Kramer (ed.), *Adaptation of Plants to Water and High Temperature Stress*. John Wiley and Sons, New York, 1980; 7-19.
7. Wery J, Silim SN, Knights EJ, Malhotra RS, Cousin R. Screening techniques and sources and tolerance to extremes of moisture and air temperature in cool season food legumes, *Euphytica*. 1994; 73:73-83.
8. Souza RP, Machado EC, Silva JAB, Lagôa AM, Silveira JAG. Photosynthetic gas exchange, chlorophyll fluorescence and some associated metabolic changes in cowpea (*Vigna unguiculata*) during water stress and recovery. *Environmental and Experimental Botany*, 2004; 51:45-56.
9. Zlatev ZS, Yordanov IT. Effects of soil drought on photosynthesis and chlorophyll fluorescence in bean plants. *Bulgarian J Plant Physiol*. 2004; 30:3-18.
10. Carmen IU, Chithra P, Huang Q, Takhistov P, Liu S, Kokini JL. *et al* Nanotechnology: a new frontier in food science. *Food Technol*. 2003; 57:24-29.
11. Nair R, Varghese SH, Nair BG, Maekawa T, Yoshida Y, Kumar DS. *et al* Nanoparticulate material delivery to plants. *Plant Sci*. 2010; 179:154-163.
12. Ball P. Natural strategies for the molecular engineer. *Nanotechnology*. 2002; 13(5):15-28.
13. Roco MC, Bainbridge WS. Converging technologies for improving human performance: nanotechnology, 2003.
14. Nel T, Xia L, Mädler NL. Toxic potential of materials at the nano level. 2006; 311:622-627.
15. Brunner TI, Wick P, Manser P, Spohn P, Grass RN, Limbach L. *et al*. *In-vitro* cytotoxicity of oxide nanoparticles: comparison to asbestos, silica and effect of particle solubility. *Environmental Science & Technology*. 2006; 40:4347-4381.
16. Matei IC, Cadar O, Roman C, Schiopu V, Synthesis and characterization of ZnO-polymer nanocomposites. *Int. J Mater. Form*, 2008, 1767-770. 10.1007/s12289-008-0288-5.
17. Abdul-Baki AA, Anderson JD. 'Vigor determination in soybean and seed multiple criteria', *Crop Sci*. 1973; 13(6):630-633.
18. Lowry OH, Poesenbrough NJ, Fal AL, Randall RJ. Protein measurement with folin phenol reagent, *J Biol. Chem*. 1951; 193:265-275.
19. Aron DI. Copper enzyme in isolated chloroplast. Polyphenol oxidase in beta vulgaris. *Plant Physiology*. 1949; 24:1-15
20. Bates LS, Waldren RP, Teare ID. *Plant & Soil*. 1973; 39:205.
21. Nel T, Xia L, Mädler NL. Toxic potential of materials at the nano level. 2006; 311:622-627.
22. Beauchamp CO, Fridovich I. Superoxide dismutase: improved assay and an assay applicable to acrylamide gels. *Anal. Biochem*. 1971; 44:276-287.
23. Gorin N, Heidema FT. Peroxidase activity in golden delicious apples as a p Theodore Roosevelt possible parameter of ripening and senescence. *J Agric. Food Chem*. 1976; 24:200-201
24. Beauchamp CO, Fridovich I. Superoxide dismutase: improved assay and an assay applicable to acrylamide gels. *Anal. Biochem*. 1971; 44:276-287.
25. Brunner TI, Wick P, Manser P, Spohn P, Grass RN, Limbach L. *et al*. *In-vitro* cytotoxicity of oxide nanoparticles: comparison to asbestos, silica and effect of particle solubility. *Environmental Science & Technology*. 2006; 40:4347-4381.
26. Efeoğlu B, Ekmekci Y, Cicek N. Physiological responses of three maize cultivars to drought stress and recovery. *South African Journal of Botany*. 2009; 75(1):34-42.
27. Morizet T, Pollucsk M, Togola D. Drought tolerance in four maize varieties (*Field Crops Abst*. 1983; 39:306-1986.
28. Thakur PS, Rai VK. Water stress effects on maize growth responses of two differentially drought sensitive maize cultivars during early stage of growth. *Indian Journal of Ecology*. 1984; 11:92-98.
29. Farooq M, Wahid A, Kobayashi N, Fujita D. Basra, Plant drought stress: effects, mechanisms and management. *Agron. Sustain. Dev*. 2009; 29:185-212.
30. Chugh V, Kaur N, Grewal MS, Gupta AK. Differential antioxidative response of tolerant and sensitive maize (*Zea mays* L.) genotypes to drought stress at reproductive stage. *Indian Journal of Biochemistry and Biophysics*. 2013; 50:150-158.
31. Shen X, Zhou Y, Duan L, Li Z, Eneji A, Li J. *et al*. Silicon effects on photosynthesis and antioxidant parameters of soybean seedlings under drought and ultraviolet-B radiation. *Journal of Plant Physiology*. 2010; 167:1248-1252.
32. Anjum SA, Wang LC, Farooq M, Hussain M, Xue LL, Zou CM. *et al*. Brassinolide application improves the drought tolerance in maize through modulation of enzymatic antioxidants and leaf gas exchange. *J Agron. Crop Sci*. 2011; 197:177-185.